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Modulation of 11β-Hydroxysteroid Dehydrogenase Isozymes by Growth Hormone and Insulin-Like Growth Factor: *In Vivo* and *In Vitro* Studies*

J. S. MOORE, J. P. MONSON, G. KALTSAS, P. PUTIGNANO, P. J. WOOD, M. C. SHEPPARD, G. M. BESSER, N. F. TAYLOR, AND P. M. STEWART†

Division of Medical Sciences, University of Birmingham, Queen Elizabeth Hospital (J.S.M., M.C.S., P.M.S.), Edgbaston, Birmingham, United Kingdom B15 2TH; the Department of Endocrinology, St. Bartholomew's Hospital (J.P.M., G.K., P.P., G.M.B.), London, United Kingdom EC1A 7BE; the Regional Endocrine Unit, Southampton University Medical School ((P.J.W.), Southampton, United Kingdom S09 4XY; and the Department of Clinical Biochemistry, King's College School of Medicine (N.F.T.), Denmark Hill, London, United Kingdom SE5 9RS

ABSTRACT

The interconversion of hormonally active cortisol (F) and inactive cortisone (E) is catalyzed by two isozymes of 11β-hydroxysteroid dehydrogenase (11βHSD), an oxo-reductase converting E to F (11βHSD1) and a dehydrogenase (11βHSD2) converting F to E. 11βHSD1 is important in mediating glucocorticoid-regulated glucose homeostasis and regional adipocyte differentiation. Earlier studies conducted with GH-deficient subjects treated with replacement GH suggested that GH may modulate 11βHSD1 activity.

In 7 acromegalic subjects withdrawing from medical therapy (Sandostatin-LAR; 20–40 mg/month for at least 12 months), GH rose from 7.1 ± 1.5 to 17.5 ± 4.3 mU/L (mean ± SE), and insulin-like growth factor I (IGF-I) rose from 43.0 ± 8.8 to 82.1 ± 13.7 nmol/L (both $P < 0.05$) 4 months after treatment. There was a significant alteration in the normal set-point of F to E interconversion toward E. The fall in the urinary tetrahydrocortisols/tetrahydocortisone ratio (THF+allo-THF/THE; 0.82 ± 0.06 to 0.60 ± 0.06; $P < 0.02$) but unaltered urinary free F/urinary free E ratio (a marker for 11βHSD2 activity) suggested that this was due to inhibition of 11βHSD1 activity. An inverse correlation between GH and the THF+allo-THF/THE ratio was observed ($r = -0.422$; $P < 0.05$). Conversely, in 12 acromegalic patients treated by transsphenoidal surgery (GH falling from 124 ± 49.2 to 29.3 ± 15.4 mU/L; $P < 0.01$), the THF+allo-THF/THE ratio rose from 0.53 ± 0.06 to 0.63 ± 0.07 ($P < 0.05$). Patients from either group who failed to demonstrate a change in GH levels showed no change in the THF+allo-THF/THE ratio.

In vitro studies conducted on cells stably transfected with either the human 11βHSD1 or 11βHSD2 complementary DNA and primary cultures of human omental adipose stromal cells expressing only the 11βHSD1 isozyme indicated a dose-dependent inhibition of 11βHSD1 oxo-reductase activity with IGF-I, but not GH. Neither IGF-I nor GH had any effect on 11βHSD2 activity.

GH, through an IGF-I-mediated effect, inhibits 11βHSD1 activity. This reduction in E to F conversion will increase the MCR of F, and care should be taken to monitor the adequacy of function of the hypothalamo-pituitary-adrenal axis in acromegalic subjects and in GH-deficient, hypopituitary patients commencing replacement GH therapy. Conversely, enhanced E to F conversion occurs with a reduction in GH levels; in liver and adipose tissue this would result in increased hepatic glucose output and visceral adiposity, suggesting that part of the phenotype currently attributable to adult GH deficiency may be an indirect consequence of its effect on tissue F metabolism via 11βHSD1 expression. (*J Clin Endocrinol Metab* **84:** 4172–4177, 1999)

TWO ISOFORMS of 11β-hydroxysteroid dehydrogenase (11βHSD) catalyze the interconversion of hormonally active cortisol (F) and inactive cortisone (E) (1, 2). Type 1 11βHSD (11βHSD1) is a NADP(H)-dependent enzyme that acts predominantly as a reductase *in vivo*, generating F from E. Expressed in liver, gonad, and adipose tissue (3, 4), it acts as a prereceptor signaling pathway for the glucocorticoid receptor. Inhibition of 11βHSD1 results in a reduction in hepatic glucose output (5), and failure to reactivate glucocorticoid in recombinant mice lacking the 11βHSD1 gene results in a reduction in stress-induced blood glucose levels (6). In adipose tissue 11βHSD1 is preferentially expressed in visceral depots (7), where it controls glucocorticoid-mediated adipocyte differentiation (8). Thus, 11βHSD1 may play a crucial role in modulating corticosteroid-regulated glucose homeostasis, energy balance, and body fat distribution.

By contrast, the high affinity, NAD-dependent dehydrogenase type 2 11βHSD enzyme (11βHSD2), protects the mineralocorticoid receptor from illicit occupancy by F (9–11). Defects in 11βHSD2 activity, either congenital in the syndrome of apparent mineralocorticoid excess (2, 12) or acquired after licorice ingestion (13), result in F-induced, mineralocorticoid hypertension. The assessment of activity of these 11βHSD isozymes in man can be inferred from a 24-h urinary steroid metabolite profile by measuring the excretion of free and A ring-reduced F and E metabolites (14, 15).

It is possible that many of the clinical features observed in patients with aberrant GH secretion may be mediated by altered 11βHSD activity. A characteristic feature of acromegaly is hypertension, which has been thought to be due to enhanced renal sodium retention (16, 17). Conversely, GH deficiency in adults (GHD) is associated with insulin resistance and visceral obesity (18). In keeping with this hypoth-

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Address all correspondence and requests for reprints to: Prof. Paul M. Stewart, M.D., F.R.C.P., Division of Medical Sciences, University of Birmingham, Queen Elizabeth Hospital, Edgbaston, Birmingham, United Kingdom B15 2TH. E-mail: p.m.stewart@bham.ac.uk.

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† Medical Research Council Senior Clinical Fellow.

esis, recent studies conducted on hypopituitary patients suggested that GH replacement therapy altered the normal setpoint of F to E conversion toward E (19–21). The data analyzing an effect of GH on 11βHSD2 are conflicting, with studies showing either no change (20) or an increase (21) in the urinary free F/urinary free E ratio, but a common finding was the inhibition of 11βHSD1 activity, *i.e.* conversion of E to F after GH replacement. These clinical studies, however, were carried out on patients with additional endocrine deficiencies; many were taking hydrocortisone replacement, which may have confounded the results. Interpretation is further complicated by the fact that net F to E conversion in hypopituitary patients *in vivo* appears to be directly related to total and regional body fat distribution (22).

The aim of this study, therefore, was to investigate the effect of GH and/or insulin-like growth factor I (IGF-I) on 11βHSD1 and 11βHSD2 activities. We have used robust *in vitro* systems known to express only 11βHSD1 or 11βHSD2. Clinically, we have studied patients with acromegaly, all of whom had intact function of the hypothalamo-pituitary-adrenal axis, affter modulation of endogenous GH/IGF-I levels.

Subjects and Methods

In vitro studies

Primary cultures of human omental adipose stromal cells. Our earlier studies have demonstrated that 11βHSD1, but not 11βHSD2, is expressed in adipose tissue (7). Omental adipose tissue was obtained from 5 patients (mean age, 73 yr) undergoing elective abdominal surgery in accordance with guidelines on the collection and use of surgical tissues obtained from the local hospital ethical committee. Stromal cells were isolated as previously reported (7, 8). Briefly, 2–4 g wet weight of adipose tissue were washed with phosphate-buffered saline containing 50,000 U penicillin and 50,000 μg streptomycin, and then digested with collagenase class 1 (2 mg/ml; Worthington Biochemical Corp., Freehold, NJ) in 1 × Hanks' Balanced Salt Solution for 45 min at 37 C. After centrifugation at 100 × *g* for 5 min, the pellet containing the stromal cells was washed with DMEM-nutrient mixture F-12 containing 15% FCS (Life Technologies, Inc., Paisley, UK). Cells were then plated overnight in the above medium, washed with 1 × Hank's Balanced Salt Solution, and grown in DMEM/F-12 containing 100 nmol/L F. Experiments were carried out on confluent cells 5–7 days later. Twenty-four hours before GH/IGF-I treatments, medium was changed to DMEM/F-12 without phenol red containing 10 μg/ml transferrin along with penicillin and streptomycin. The cells were then treated for 48 h with 1, 10, or 100 ng/ml human recombinant GH (Pharmacia & Upjohn, Inc., Stockholm, Sweden) or 1, 10, or 100 ng/ml human recombinant IGF-I (Sigma Chemical Co., Poole, UK). In each case triplicate assays were run.

Stably transfected fetal kidney cells. HEK 293 cells are derived from human fetal kidney, which is known to express both GH (23) and IGF-I (24) receptors. 293 cells (devoid of endogenous 11βHSD) were stably transfected with either human 11βHSD type 1 complementary DNA (cDNA; 293T1) or human 11βHSD type 2 cDNA (293T2) as previously reported (25). The 11βHSD cDNAs were donated by Drs. White and Krozowski, respectively (3, 11). Cells were grown in MEM (Life Technologies, Inc.) containing 10% FCS and geneticin until confluence. Twenty-four hours before treatments, medium was changed to serum-free DMEM without phenol red containing 5 ml MEM nonessential amino acids and 150 mg glutamine. Cells were then treated with 1, 10, or 100 ng/ml GH or IGF-I for 8, 24, and 48 h. Each experiment was repeated four times in triplicate.

11βHSD enzyme assays

11βHSD activity was assayed by incubating intact cells with either 250 nmol/L E (omental and 293T1 cells) or 50 nmol/L F (293T2 cells) with appropriate tracer for 4 or 2 h, respectively. The steroid concentrations used were based on the substrate affinities of the 11βHSD1 and two enzymes and ensured first order enzyme kinetics (26). After incubation, steroids were extracted using dichloromethane and then separated using a mobile phase consisting of ethanol and chloroform (8:92) on a TLC plate (Fluka, UK). The fractional conversion of E to F or F to E was calculated after scanning the TLC plate on a Bioscan Autochanger 3000 system (Lablogic, Sheffield, UK).

Cellular protein levels in each incubate was calculated using the Bradford method (27), and enzyme activities were expressed as picomoles per mg protein/h (mean ± SE). Statistical analysis was undertaken using Student's paired *t* test.

Clinical studies

Effect of reducing GH on 11βHSD activity. Twelve patients with active acromegaly (seven women and five men; mean age, 43 yr; range, 22–60 yr) were evaluated before and after transsphenoidal surgery. In each case detailed testing of anterior pituitary function indicated normal adrenal, thyroid, and gonadal function when 11βHSD activity was analyzed pre- and postoperatively. The time of testing after pituitary surgery varied between 8–16 weeks. GH was measured using an in-house immunoradiometric assay developed by the reference laboratory (NETRIA) based at St. Bartholomew's Hospital (London, UK). IGF-I was measured as previously reported (28).

Effect of increasing GH on 11βHSD activity. Seven patients with acromegaly (two men and five women; mean age, 49 yr) were evaluated after withdrawal of Sandostatin-LAR (Novartis Pharmaceuticals; 20–30 mg every 28 days) (29). Three of the seven patients had microadenomas, and all patients had intact hypothalamo-pituitary-adrenal reserve (as reflected by a serum F >550 nmol/L 30 min after im Synacthen). One patient was receiving replacement T_4, but thyroid function tests in every patient were normal and did not change across withdrawal of Sandostatin-LAR. All patients had received a minimum of 12 months of therapy with Sandostatin LAR; the clinical and biochemical responses to withdrawal of therapy and the details of the GH and IGF-I assays employed for this study have been recently published (30). Patients were investigated at 4-week intervals after withdrawal of Sandostatin-LAR for a total of 16 weeks.

Both clinical studies had the approval of the local hospital ethical committees. In both studies, IGF-I was measured basally, and GH was determined at hourly intervals for 5 h. Patients completed a 24-h urine collection, which was assayed for F metabolites as previously described(20), using gas chromatography/mass spectrometry. The tetrahydrocortisols/tetrahydrocortisone (THF+allo-THF/THE), 5α-THF/5β-THF, and urinary free F (UFF)/urinary free E (UFE) ratios were calculated together with total F metabolite excretion (THF+allo-THF+THE+cortols+cortolones+UFF) and are expressed as the mean ± SE. In each case statistical analysis was undertaken using Student's paired *t* test.

Results

In vitro studies

Incubation of both primary cultures of omental adipose stromal cells (ASC) and 293T1 cells with IGF-I resulted in a dose-dependent inhibition of reductase activity (E to F conversion). In the 293T1 cells, no effect was evident after 8 h of incubation with IGF-I, and some inhibition was observed after 24 h of incubation (data not shown), but the most marked effect was observed at 48 h of incubation (Fig. 1). At this time point, inhibition was observed only with 10 and 100 ng/mL IGF-I; no effect of 1 ng/mL IGF-I was observed. In contrast, GH had no effect on 11βHSD1 activity, even at a concentration of 100 ng/mL, at incubation times of 48 h.

The effect on 11βHSD1 was specific in that neither IGF-I nor GH, had any effect on 11βHSD2 activity in 293T2 cells. Similar negative results were obtained using the human colon cell line, SW620 cells, which expresses 11βHSD2 but not 11βHSD1 (data not shown).

For the omental ASC experiments, no differentiation from

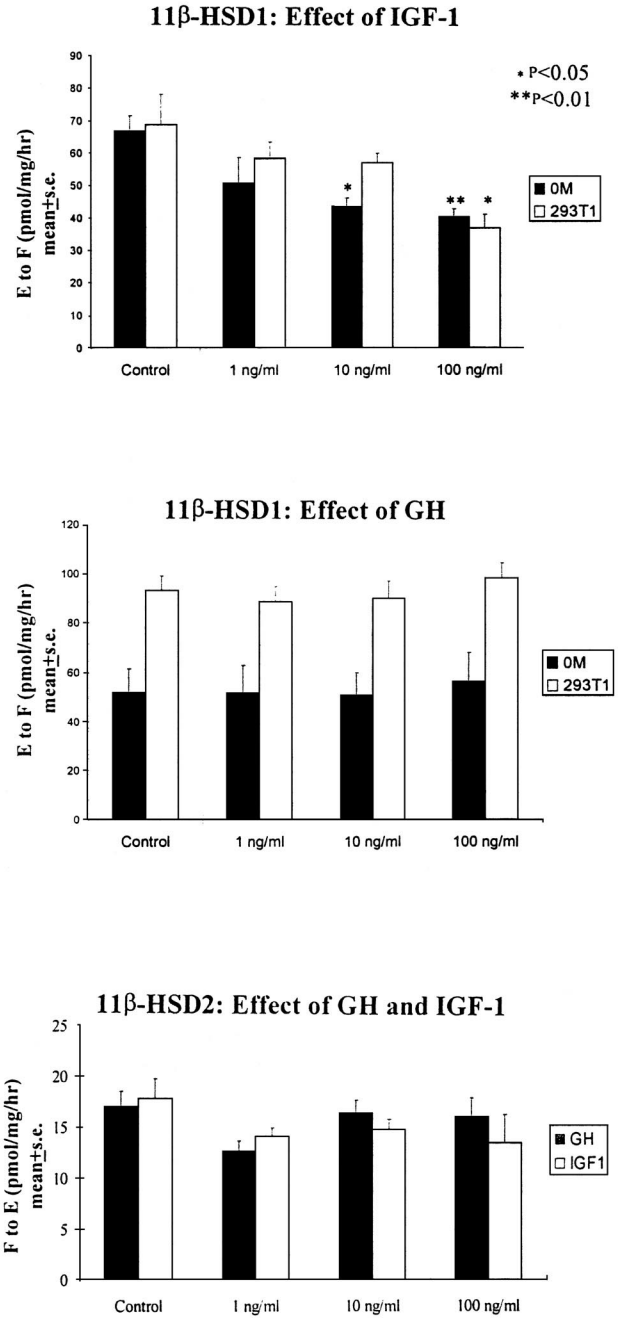

FIG. 1. The effect of GH/IGF-I on 11βHSD activity. *Top panel*, The effects of increasing doses of IGF-I on 11βHSD1 reductase activity in either primary cultures of human omental adipose stromal cells (om) or cultures of human fetal kidney 293 cells stably transfected with 11βHSD1 (293T1). *Middle panel*, As above, but with cells incubated with GH. *Bottom panel*, The effects of GH and IGF-I on 11βHSD2 activity in fetal kidney 293 cells stably transfected with 11βHSD2. All incubations were performed for 48 h. Each experiment was conducted in triplicate and represents the mean of four separate assays.

ASC to adipocytes was observed after incubation with GH or IGF-I for 48 h (longer incubation times with putative differentiation agents are required to induce differentiation of these primary cultures) (8). Furthermore, in all of the experiments, at least over this 48-h period, there did not appear to be a trophic effect of either GH or IGF-I as determined by cellular protein assays.

Clinical studies

The influences of surgery on mean serum GH, IGF-I, THF+alloTHF/THE, alloTHF/THF, and total F metabolite excretion are shown in Table 1. Significant increments in the ratios of THF+alloTHF/THE were observed for the group as a whole ($P = 0.045$; Fig. 2) and were particularly evident in those patients who demonstrated a substantial reduction in serum GH as a result of surgery. In four patients there was no discernible change in the THF+alloTHF/THE, and of these patients, three showed no reduction in GH and/or IGF-I after surgery (patients 4, 8, and 9 in Table 1). There was no correlation between either GH or IGF-I and the THF+alloTHF/THE ratio. There was no change in the ratio of allo-THF/THF or in total F metabolite excretion after surgery.

In seven acromegalic patients withdrawing from Sandostatin-LAR, there was a significant increase in GH and IGF-I levels, as recently reported. Thus, GH rose from a baseline value of 7.1 ± 1.5 mU/L (mean ± SE; n = 7) to 17.0 ± 4.3 mU/L 16 weeks after Sandostatin-LAR, and IGF-I rose from 43.0 ± 8.8 to 82.1 ± 13.7 nmol/L. This was associated with a significant reduction in the THF+alloTHF/THE ratio from 0.82 ± 0.06 to 0.60 ± 0.06 ($P = 0.02$; Table 2 and Fig. 2). The UFF/UFE ratio, however, did not change (0.64 ± 0.05 and 0.61 ± 0.04, respectively). GH did not rise in one of the seven patients, remaining between 4.5–5.5 mU/L; this was the only patient in whom no change in the THF+alloTHF/THE ratio was observed (0.62, 0.63, 0.60, and 0.60, respectively, 4, 8, 12, and 16 weeks after Sandostatin-LAR). No changes in the allo-THF/THF ratio or excretion of total F metabolites were seen.

A significant inverse correlation between GH and the THF+allo-THF/THE ratio was observed ($r = -0.422$; $P < 0.05$; $n = 28$).

Discussion

Our clinical studies have demonstrated a marked change in 11βHSD activity in patients with aberrant GH secretion. Patients with untreated acromegaly demonstrate a reduction in the THF+allo-THF/THE ratio, and this increases when GH (and consequently IGF-I) levels are reduced. Conversely, when F metabolism is serially analyzed in a cohort of treated acromegalic patients withdrawing from the effective depot, long acting somatostatin analog, Sandostatin LAR, the THF+allo-THF/THE ratio decreases.

Two isozymes of 11βHSD are known to coordinate the interconversion of F and E in human tissues: 11βHSD1, an oxo-reductase expressed in liver, gonad, adipose tissue, and central nervous system tissues, and 11βHSD2, a dehydrogenase expressed in the mineralocorticoid target tissues, kidney, and colon. This alteration in the normal set-point of F to E conversion toward E with increasing GH and IGF-I levels could be secondary to either enhanced 11βHSD2 activity or inhibition of 11βHSD1 activity. *In vivo* the urinary THF+allo-THF/THE ratio probably reflects "global" 11βHSD activity, with contributions from 11βHSD1 and 11βHSD2. Indeed,

TABLE 1. Serum GH, IGF-I, (THF+alloTHF)/THE,allo-THF/THF, and total cortisol metabolites in 12 patients with active acromegaly, before (i) and after (ii) transsphenoidal surgery

Patient no.	GH (mU/L) (i)	GH (mU/L) (ii)	IGF-I (ng/mL) (i)	IGF-I (ng/mL) (ii)	THF+allo THF/THE (i)	THF+allo THF/THE (ii)	AlloTHF/THF (i)	AlloTHF/THF (ii)	Total cortisol metabolites (μg/24 h) (i)	Total cortisol metabolites (μg/24 h) (ii)
1	87.6	42	995		0.38	0.72	0.38	0.27	22,503	11,564
2	72	15.6	685	276	0.83	1.13	0.59	0.54	6,051	11,299
3	28	3	386	148	0.78	0.64	0.51	0.95	4,754	3,907
4	15	20.6	547	487	0.77	0.75	0.31	0.33	9,488	16,226
5	34	5.1	526	228	0.52	0.51	0.17	0.19	4,623	4,933
6	92.9	1.8	731	207	0.30	0.56	0.48	0.22	10,889	7,425
7	40.4	6.6	767	239	0.65	0.82	0.45	0.59	9,509	9,241
8	570	190	1413	1441	0.54	0.46	0.9	1.08	17,722	19,047
9	52.5	54	697		0.34	0.32	0.19	0.28	14,117	4,917
10	102	2.4	942		0.69	0.79	0.92	0.69	7,224	16,719
11	21.2	2.3	435	224	0.31	0.49	0.48	0.41	7,883	12,294
12	375	8.0	1.019	378	0.26	0.40	0.2	0.18	8,029	12,716
Mean ± SEM	124.2 ± 49.2	29.3 ± 15.4[a]	723.2 ± 107.6	403.1 ± 134.1[a]	0.53 ± 0.06	0.63 ± 0.07[b]	0.47 ± 0.07	0.48 ± 0.09	10,233 ± 1561	10,857 ± 1429

[a] $P < 0.01$, i *vs.* ii.
[b] $P < 0.05$, i *vs.* ii.

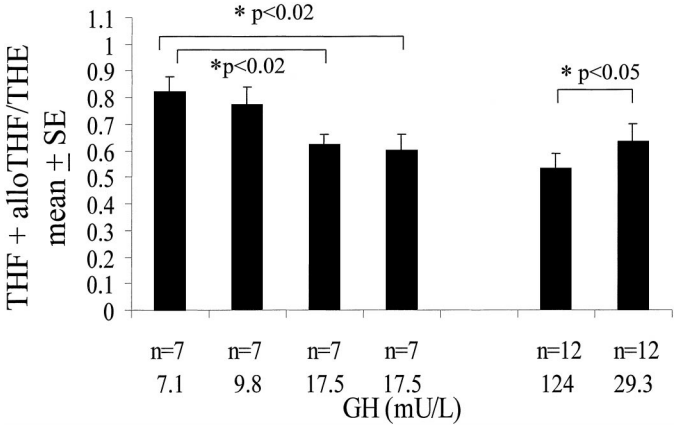

FIG. 2. The effect of altered GH secretion on the urinary THF+allo-THF/THE ratio in patients with acromegaly. On the *left*, 7 patients with acromegaly withdrew from Sandostatin-LAR, and the urinary THF+allo-THF/THE ratio was measured at 4-week intervals for a total of 16 weeks after the last injection. In the groups as a whole, GH rose from 7.1 to 17.5 mU/L, IGF-I rose from 43.0 to 82.1 nmol/L (both $P < 0.05$), and the THF+allo-THF/THE ratio fell. On the *right*, 12 newly diagnosed patients with acromegaly were treated with transsphenoidal hypophysectomy. Mean GH fell from 124 to 29.3 mU/L, and IGF-I fell in the group as a whole; this was associated with a significant increase in the THF+allo-THF/THE ratio.

this ratio is grossly elevated in patients with apparent mineralocorticoid excess due to inactivating mutations in the human gene encoding 11βHSD2 (2). [Such patients have completely normal 11βHSD1 activity (31).] Similarly, the ratio is equally deranged, but in the opposite direction, in patients with the syndrome of apparent E reductase deficiency, which is thought to be due to loss of 11βHSD1, but not 11βHSD2, activity (32). The UFF/UFE ratio may be a more accurate marker of 11βHSD2 activity; UFE excretion is 2- to 3-fold higher than UFF, reflecting renal conversion of F to E by 11βHSD2 (15). Thus, our observation of a reduction in the THF+allo-THF/THE ratio, but a normal UFF/UFE ratio, with increasing GH is highly suggestive of inhibition of 11βHSD1 reductase activity. The lack of change in the UFF/UFE ratio after these alterations in GH/IGF-I status is in keeping with our earlier data (20) and suggests that inhibition of renal 11βHSD2 activity is not implicated in the pathogenesis of sodium retention observed in patients with acromegaly.

This change in the pattern of F metabolism, however, may have clinical ramifications for glucocorticoid status in both acromegalic and hypopituitary, GH-deficient, patients. By inhibiting 11βHSD1, GH would effectively increase the MCR of F. As a result we recommend close monitoring of the pituitary-adrenal axis both in patients starting GH replacement therapy and in untreated acromegalic patients. Secondary adrenal failure may be precipitated in such situations, and patients with documented hypothalamo-pituitary-adrenal axis deficiency already taking replacement hydrocortisone may require an escalation of replacement dose.

Our *in vitro* studies endorse our clinical studies. The effect of GH could be due to a direct effect of GH itself or could be mediated by IGF-I. GH, however, had no effect on 11βHSD1 (or 11βHSD2) activity, but IGF-I caused a dose-dependant inhibition of 11βHSD1 reductase activity. No alteration in 11βHSD2 dehydrogenase activity after incubation with IGF-I was observed. These data are in keeping with some earlier *in vitro* studies, but not others. Thus, in rodents, the sexual dimorphic expression of hepatic 11βHSD1 is thought to be secondary to sex-specific changes in GH secretion (33). The higher expression of 11βHSD1 activity in male rat liver, for example, can be reduced to female values by a continuous female pattern of GH administration. Such inhibition of 11βHSD1 could, of course, in an intact animal, be mediated by IGF-I. By contrast, GH itself has been shown to inhibit 11βHSD1 activity in cultured rat hepatocytes (34), although other studies have failed to confirm these findings (35).

In addition to cells stably transfected with 11βHSD1, our *in vitro* studies used primary cultures of omental adipose stromal cells, and these observations may be of considerable relevance. For reasons that are still unclear, 11βHSD1

TABLE 2. Serum GH and IGF-I and urinary cortisol metabolites in seven acromegalic patients withdrawing from Sandostatin-LAR (S-LAR)

Weeks post-S-LAR	GH (mU/L)	IGF-I (nmol/L)	THF+allo-THF/THE	Allo-THF/THF	UFF/UFE	Total F metabolites (μg/24 h)
4	7.1 ± 1.5	43.0 ± 8.8	0.82 ± 0.06	0.44 ± 0.09	0.64 ± 0.05	8021 ± 1373
8	9.8 ± 2.6		0.77 ± 0.07	0.40 ± 0.08	0.61 ± 0.05	7284 ± 694
12	17.5 ± 4.0[a]		0.62 ± 0.04[b]	0.46 ± 0.11	0.62 ± 0.07	7879 ± 959
16	17.5 ± 4.3[a]	82.1 ± 13.7[a]	0.60 ± 0.06[b]	0.44 ± 0.09	0.61 ± 0.04	8131 ± 1506

[a] $P < 0.05$.
[b] $P < 0.02$.

is preferentially expressed in omental compared to sc adipose depots (7), where it plays a crucial role in dictating glucocorticoid-induced adipocyte differentiation (8). This may be one mechanism explaining the association of glucocorticoids with visceral, as distinct from generalized, obesity. The observation that IGF-I can modulate this process may have ramifications for patients with adult GH deficiency. Earlier studies suggested that E to F metabolism (*i.e.* 11βHSD1 activity) was enhanced in GH-deficient patients with hypopituitarism (19); these data as well as data showing the opposite in patients with GH excess suggest that GH deficiency, through its effect on 11βHSD1, would increase omental adiposity. Conversely, GH treatment to such a cohort, again through an action on 11βHSD1, would be expected to reverse this process. Similarly, within the liver, another principal site of 11βHSD1 expression, increased GH might be expected to inhibit hepatic glucocorticoid levels, thereby reducing gluconeogenesis and hepatic glucose output (5, 6). Thus, it is exciting to speculate that many of the clinical features currently attributed to adult GH deficiency, such as central obesity and insulin resistance (18), may in part be mediated by glucocorticoids through IGF-I modulation of tissue 11βHSD1 activity.

References

1. **Stewart PM.** 1996 11β-Hydoxysteroid dehydrogenase: implications for clinical medicine Clin Endocrinol (Oxf). 44:493–499.
2. **White PC, Mune T, Agarwal AK.** 1997 11β-hydroxysteroid dehydrogenase and the syndrome of apparent mineralocorticoid excess. Endocr Rev. 18:135–156.
3. **Tannin GM, Agarwal AK, Monder C, New MI, White PC.** 1991 The human gene for 11β-hydroxysteroid dehydrogenase. Structure, tissue distribution, and chromosomal localization. J Biol Chem. 266:16653–16658.
4. **Ricketts ML, Verhaeg JM, Bujalska I, Howie A, Rainey WE, Stewart PM.** 1998 Immunohistochemical localization of type 1 11β-hydroxysteroid dehydrogenase in human tissues. J Clin Endocrinol Metab. 83:1325–1335.
5. **Walker BR, Connacher AA, Lindsay RM, Webb DJ, Edwards CRW.** 1995 Carbenoxolone increases hepatic insulin sensitivity in man: a novel role for 11-oxosteroid reductase in enhancing glucocorticoid receptor activation. J Clin Endocrinol Metab. 80:3155–3159.
6. **Kotelevtsev YV, Holmes MC, Burchell A, et al.** 1997 11β-Hydroxysteroid dehydrogenase type 1 knockout mice show attenuated glucocorticoid-inducible responses and resist hyperglycaemia on obesity or stress. Proc Natl Acad Sci USA. 94:14924–14929.
7. **Bujalska IJ, Kumar S, Stewart PM.** 1997 Does central obesity reflect "Cushing's disease of the omentum?" Lancet. 349:1210–1213.
8. **Bujalska IJ, Hewison M, Kumar S, Stewart PM.** 1999 Differentiation of omental adipose stromal cells: role of glucocorticoids and 11β-hydroxysteroid dehydrogenase. Endocrinology. 140:3188–3196.
9. **Edwards CRW, Stewart PM, Burt D, et al.** 1988 Localisation of 11β-hydroxysteroid dehydrogenase-tissue specific protector of the mineralocorticoid receptor. Lancet. 2:986–989.
10. **Funder JW, Pearce PT, Smith R, Smith AI.** 1988 Mineralocorticoid action: target tissue specificity is enzyme, not receptor, mediated. Science. 242:583–585.
11. **Albiston AL, Obeyesekere VR, Smith RE, Krozowski ZS.** 1994 Cloning and tissue distribution of the human 11β-hydroxysteroid dehydrogenase type 2 enzyme. Mol Cell Endocrinol. 105:R11–R17.
12. **Stewart PM, Krozowski ZS, Gupta A, Milford DV, Howie AJ, Sheppard MC, Whorwood CB.** 1996 Hypertension in the syndrome of apparent mineralocorticoid excess due to mutation of the 11β-hydroxysteroid dehydrogenase type 2 gene. Lancet. 347:88–91.
13. **Stewart PM, Wallace AM, Valentino R, Burt D, Shackleton CHL, Edwards CRW.** 1987 Mineralocorticoid activity of liquorice: 11β-hydroxysteroid dehydrogenase deficiency comes of age. Lancet. 2:821–824.
14. **Shackleton CHL.** 1993 Mass spectrometry in the diagnosis of steroid-related disorders and hypertension research. J Steroid Biochem Mol Biol. 45:127–140.
15. **Palermo M, Shackleton CHL, Mantero F, Stewart PM.** 1996 Urinary free cortisone and the assessment of 11β-hydroxysteroid dehydrogenase activity in man. Clin Endocrinol (Oxf). 45:605–611.
16. **Bengtsson B-Å, Brummer R-JM, Edén S, Bosaeus I.** 1989 Body composition in acromegaly. Clin Endocrinol (Oxf). 30:121–130.
17. **Sacca L, Cittadini A, Fazio S.** 1994 Growth hormone and the heart. Endocr Rev. 15:555–573.
18. **Deboer H, Blok, GJ, Vanderveen EA.** 1995 Clinical aspects of growth hormone deficiency in adults. Endocr Rev. 16:63–86.
19. **Weaver JU, Thaventhiran L, Noonan K, Burrin JM, Norman M, Taylor NF, Monson JP.** 1994 The effect of growth hormone replacement on cortisol metabolism and glucocorticoid sensitivity in hypopituitary adults. Clin Endocrinol (Oxf). 41:639–648.
20. **Gelding SV, Taylor NF, Wood PJ, Noonan K, Weaver JU, Wood DF, Monson JP.** 1998 The effect of growth hormone replacement therapy on cortisol-cortisone interconversion in hypopituitary adults: evidence for growth hormone modulation of extrarenal 11β-hydroxysteroid dehydrogenase activity. Clin Endocrinol (Oxf). 48:153–162.
21. **Walker BR, Andrew R, MacLeod KM, Padfield PL.** 1998 Growth hormone replacement inhibits renal and hepatic 11β-hydroxysteroid dehydrogenases in ACTH deficient patients. Clin Endocrinol (Oxf). 49:257–263.
22. **Weaver JU, Taylor NF, Monson JP, Wood PJ, Kelly WF.** 1998 Sexual dimorphism in 11β hydroxysteroid dehydrogenase activity and its relation to fat distribution and insulin sensitivity; a study in hypopituitary subjects. Clin Endocrinol (Oxf). 49:13–20.
23. **Hill DJ, Riley SC, Bassett NS, Waters MJ.** 1992 Localization of the growth hormone receptor, identified by immunocytochemistry, in second trimester human fetal tissues and in placenta throughout gestation. J Clin Endocrinol Metab, 75:646–650.
24. **Grone HJ, Neumann P, Fuchs E.** 1992 Localization and characterization of IGF-1 receptors in fetal and adult human kidneys. Miner Electrolyte Metab. 18:256–263.
25. **Bujalska I, Shimojo M, Howie A, Stewart PM.** 1997 Human 11β-hydroxysteroid dehydrogenase: studies on the stably transfected isoforms and localization of the type 2 isozyme within renal tissue. Steroids. 77:77–82.
26. **Stewart PM, Murry BA, Mason JI.** 1994 Human kidney 11β-hydroxysteroid dehydrogenase is a high affinity NAD^+-dependent enzyme and differs from the cloned "type I" isoform. J Clin Endocrinol Metab. 79:480–484.
27. **Bradford MM.** 1976 A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principal of protein dye binding. Anal Biochem. 72:248–252.
28. **Morrell DJ, Dadi H, More J, et al.** 1989 A monoclonal antibody to human insulin-like growth factor-I: characterisation, use in radioimmunoassay and effect on the biological activities of the growth factor. J Mol Endocrinol. 2:201–206.
29. **Stewart PM, Kane K, Stewart SE, Lancranjan I, Sheppard MC.** 1995 Depot long-acting somatosatin analog (Sandostatin-LAR) is an effective treatment for acromegaly. J Clin Endocrinol Metab. 80:3267–3272.
30. **Stewart PM, Stewart SE, Clark PMS. Sheppard MC.** 1999 Clinical and biochemical response following withdrawal of a long-acting, depot injection form of octreotide (Sandostatin LAR). Clin Endocrinol (Oxf). 50:295–300.
31. **Stewart PM, Corrie JET, Shackleton CHL, Edwards CRW.** 1988 Syndrome of apparent mineralocorticoid excess: A defect in the cortisol-cortisone shuttle. J Clin Invest. 82:340–349.
32. **Phillipov G., Palermo M, Shackleton CHL.** 1996 Apparent cortisone reductase

deficiency: a unique form of hypercortisolism. J Clin Endocrinol Metab. 81:3855–3860.
33. **Low SC, Chapman KE, Edwards C, Wells T, Robinson IC, Seckl JR.** 1994 Sexual dimorphism of hepatic 11β-hydroxysteroid dehydrogenase in the rat: the role of growth hormone patterns. J Endocrinol. 143:541–548.
34. **Liu YJ, Nakagawa Y, Nasuda K, Saegusa H, Igarashi Y.** 1996 Effect of growth hormone, insulin and dexamethasone on 11β-hydroxysteroid dehydrogenase activity on a primary culture of rat hepatocytes. Life Sci. 59:227–234.
35. **Jamieson P, Chapman KE, Edwards CRW, Seckl JR.** 1995 11β-hydroxysteroid dehydrogenase is an exclusive 11β-reductase in primary cultures of rat hepatocytes: effect of physicochemical and hormonal manipulations. Endocrinology. 136:4754–4761.

Erratum

In the article "Cortisol, androstenedione (A4), dehydroepiandrosterone sulphate (DHEAS), and 17 hydroxyprogesterone (17OHP) responses to low doses of (1–24)ACTH" by Nicola Bridges *et al.* (*The Journal of Clinical Endocrinology & Metabolism* **83:** 3750–3753), the author would like to make the following correction to the text.

Figure 1, a and c, was omitted. In Figure 2, the bars described as *solid* are horizontally hatched (▨). The figures and correct legends appear here.

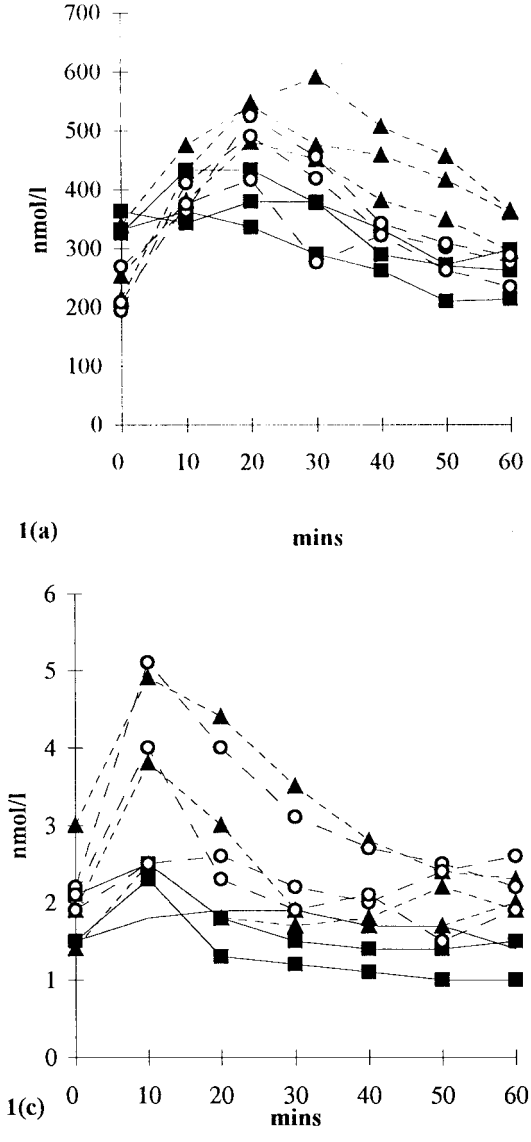

FIG. 1. Individual sampling results from one subject following administration of (1-24)ACTH in doses of 125 ng/m^2 (closed boxes and solid lines), 250 ng/m^2 (closed triangles and dotted lines) and 500 ng/m^2 (open circles and dashed lines). a) cortisol, c) 17-OHP.

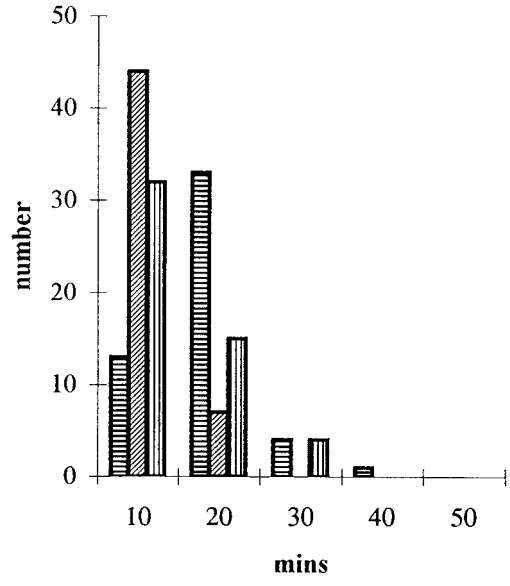

FIG. 2. Sample time of peak hormone values. Cumulative frequencies for the sample time at which peak hormone levels occurred for cortisol (horizontal lines), 17-OHP (diagonal lines) and A4 (vertical lines).